

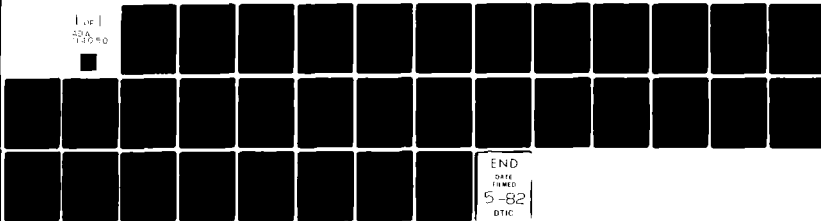
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THE USE OF GENETIC MECHANISMS AND BEHAVIORAL CHARACTERISTICS TO--ETC(U)  
MAR 82 M H ROSS, D G COCHRAN N00014-77-C-0246

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20. ABSTRACT (Continue on reverse side if necessary and identify by block number) The German cockroach has been the subject of many studies, yet there is a dearth of knowledge concerning the behavior and growth of natural populations. The work conducted under the present Contract focuses on this problem. Since natural populations consist of all age classes, laboratory and "field" experiments utilizing mixed age groups are being carried out. The results are providing basic biological data as well as information requisite to an improved technique for the use of sterile males as part of an integrated approach to		

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cockroach control.

Research summarized below was conducted during the 2nd year of the present Contract. It is presented according to specific objectives as stated in the proposal for the present phase of these studies. Objectives #1 and 4 are omitted because the first (#1) was completed last year and the other (#4) is planned for the present year of this Contract.

Objective 2 - Within-harborage behavior. A laboratory experiment was designed to study inter- and intra-component aggregation relationships of a mixed age group within a simulated harborage. The harborage area consisted of side-by-side shelters that partitioned the area into 4 homogenous sections. The only component (age class) which tended to remain outside the harborage was small nymphs (30%). Application of a dispersion statistic to the cockroaches within the harborage showed the reproductive state and density of adult females affected the aggregation of other components, as well as the aggregation of the total population. The only experiment in which males showed significant aggregation with themselves and other components was when there were low numbers of non-gravid females. The effect of increased female density was apparently reversed in groups which had non-gravid as compared to those with gravid females.

Objective 3 - Behavior and growth of cockroach populations. The aims of this objective were to study dispersion from established groups, mixing between groups in closely co-located harborages, group establishment, and the effects of the immediate surroundings (food, water & harborage) on group development. Data were gathered from a "field" experiment on an inactive ship. Eleven replicate groups of wild-type were established at sites where food, water, and harborage were provided. A summer-long trapping program was conducted in which traps were placed at "main" sites (location of food, water & releases) and at various distances and positions from the main sites. Most catch was at the main sites, although as groups at these sites developed, catch in more distantly located traps increased. Solid walls and open spaces acted as barriers to movement. Cockroaches moved freely through decayed walls. Within 3 mo (one generation), the overall wild-type population increased by 18.6x. However, development within the various groups differed according to the immediate surroundings. Removal of food and water from a favorable site resulted in a sharp drop in numbers at that site and in increased catch at neighboring trapping sites. Most movement away from the site was towards established groups. Increased movement increased the % catch. By late summer, a large group had become established in a decayed wall. Both wild-type and mutants from groups at less favorable sites were apparently attracted towards this large group. However, groups which were apparently at high density for their particular harborage seemed to have a repellent effect. This was seen in mutant as well as wild-type cockroaches. By mid instars, each of 3 successive sets of black-body nymphs (hatch of successive egg cases) moved a greater distance from their initial harborage than the preceding set. In general, least movement was by gravid females and small nymphs; most away-from-harborage movement was by mid to late instar nymphs and new adults. Age class structures as seen from trapping at main and peripheral sites throughout the summer and from final collections (carton, i.e. harborage; last set of traps, and collection at time of insecticide treatment) are currently being analyzed. Also, a higher proportion of non-gravid than gravid females were found in traps than in the carton-type harborage, supporting the laboratory experiment noted below.

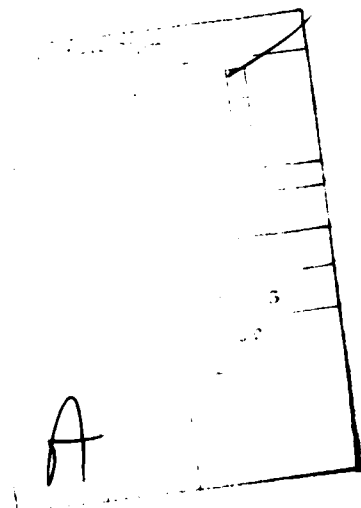
Abstract (cont.):

Objective 5 - Foraging habits. Adult females have been studied over the course of their reproductive cycle in order to determine their needs for food and water. The consumption of food and water was recorded on a daily basis for individually caged adults. The data were summarized according to certain landmark events such as maturation, mating, appearance of an egg case, and egg case hatch.

It was found that adult females eat and drink on a cyclic basis. Large amounts of food and water are consumed for several days prior to formation of an egg case. Both food and water consumption drop dramatically at the production of an egg case. Adult females eat and drink sparingly while carrying the egg case, but repeat the cycle of extensive feeding and drinking each time an egg case is dropped.



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OFFICE OF NAVAL RESEARCH

Contract #N00014-77C-0246

Task No. NR 205-028

ANNUAL REPORT NO. 4

The Use of Genetic Mechanisms and Behavioral Characteristics to  
Control Natural Populations of the German Cockroach

by

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March 1982

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The research conducted during the past year was part of a three year study designed to gain a more in depth understanding of the behavior and dynamics of cockroach populations. This is important to both a more effective use of current control techniques and the application of new approaches, such as the incorporation of a "sterile" male release into a control program. The results of a pilot test in genetic control, supported by the first years of this Contract, called attention to a dearth of knowledge concerning natural populations. It has been noted in the literature that the German cockroach is characterized by highly variable behavior, but what factors underlie this variation?

Laboratory and field research under this Contract is beginning to provide answers to the above query. In the laboratory, experiments have revealed hitherto unsuspected effects of female density and reproductive state on within-harborage aggregation behavior (Obj. 2). Other laboratory experiments show cyclic feeding and drinking behavior of females (Obj. 5). An experiment on an inactive ship has resulted in extensive data on movement, mixing between groups, favored harborages, rates of population growth, factors affecting catch in a trapping program, and other aspects of population growth and behavior (Obj. 3). These data are summarized below according to the appropriate objective. Objectives not included in this year's work are: Obj. 1 - completed earlier (sterile male experiment) and Obj. 4 - an experiment on insecticide induced dispersal, planned for the summer of 1982.

#### Objective 2 - Behavior within harborages

A laboratory study of aggregation behavior of mixed populations of B. germanica was conducted by the graduate student supported by this Contract, Brian L. Bret (M.S. thesis research, directed by M. H. Ross).

The experiment was conducted in glass aquaria measuring 32 cm x 22 cm x 21 cm. Harborage was provided by four side by side shelters (10 cm x 4 cm x 1 cm), open at each end, that were made of black construction paper. These partitioned the harborage area into four homogenous sections. A small, mixed population of wild-type B. germanica was released into each harborage. A basic group of 4 females, 4 males, 16 medium-sized (3rd to 4th stage) and 16 small nymphs (1st to 2nd stage) was used. Throughout the experiment, four or multiples thereof were used since 4 shelters were available. Two variables were introduced into the experiment: female reproductive stage and female density. The former consisted of females without oothecae, females with immature oothecae (stage I of embryonic development), and females with more advanced oothecae (stage IX to X). Differing female densities consisted of 4, 8, and 16 females. From 5 to 8 replicates of each of 9 variations were performed (3 different reproductive states at each of 3 females densities).

Each experiment was run for two days. Food and water were provided during the first day while the cockroaches became adjusted to their surroundings. On the 2nd day, these resources were removed in order to prevent their use as a harborage. At the end of the 2nd day, the shelters were sealed, cockroaches within were anesthetized with CO<sub>2</sub> and censused. New harborages were supplied for each replicate. The aquaria were washed thoroughly after each use.

The statistical analysis was done with the help of Dr. Golde Holtzman, VPI Department of Statistics. A new application of Morista's dispersion index was devised:

$$(\delta = \frac{R}{D} \frac{\# \text{ of pairs of individuals in same shelter}}{\# \text{ of possible pairs of individuals in population}}) \text{ (Holtzman, Bret and Ross, unpubl.)}$$

The  $\delta$  values (formerly referred to as "ro") were used to show statistically significant differences in aggregation and also distinctive trends towards more intense or weaker aggregation which were clearly of biological significance. A value of 0.25 indicated random dispersal. Higher values showed various degrees of aggregation, of the population as a whole and of inter- and intra-component aggregation.

Fig. 1 shows the percentage of each age class (component) of the populations according to four "re-numbered" shelters. The cockroaches tended to aggregate in one shelter, but since this was not the same shelter in the various experiments, presenting the data according to the actual physical position of each shelter gives a misleading picture of the distribution. Therefore, the data are presented with the shelter containing the largest number of females (and also of the other components) numbered "1", that with the 2nd largest no. as "2", etc. Small nymphs were the only component of which many were found outside the shelter. These apparently sought narrower hiding places, clustering in the corner of the aquaria or in cracks between the shelters. However, within the shelter, their aggregation was clearly related to that of other components, especially females (see below). Certain general aggregation characteristics, revealed in more detail by the  $\delta$  value calculations, are evident in Fig. 1. For example, males tended to disperse more than the other components of the population. Females generally aggregated with themselves, if not with other components. It is noteworthy that females were rarely found outside the shelters.

Table 1 summarizes the results according to the density of the females. Each density includes experiments with females in the three reproductive states. This provided sufficient data to demonstrate statistically significant differences, but tended to obscure certain important inter-relationships between density and reproductive state that emerged when each individual combination of the two variables was considered (see below). Table 1 shows an unusual aggregation pattern at 4♀: all components aggregated with themselves and with other components, a situation unique in that the males were among those showing significant aggregation. At 8♀, the population was more widely spread, due largely to lessened aggregation of females and males, both with themselves and other components. At 16♀, overall aggregation increased, with a  $\delta$  value similar to that at 4♀ (.44). The underlying relationships differed from those at 4♀ in that they reflected intense aggregation among nymphs, random dispersal among males, with females aggregating with themselves, medium nymphs, highly aggregating with small nymphs, but with no male-female aggregation. It will be shown below that strong aggregation at 4♀ was associated with one of the reproductive states (absence of an egg case). Otherwise, the overall data on female density would show increased aggregation with increased female density.

Table 2 summarizes the results according to the reproductive state of the females (A - no egg case; B - immature egg case; C - advanced egg case). Those without egg cases showed little tendency to aggregate with other components, and they dispersed randomly with other females. The latter differed markedly from the results of other experiments summarized in both Tables 1 and 2. Populations with gravid females (B and C) aggregated more strongly, especially those with immature oothecae (B). This relationship is more clearcut when the data are broken down according to the 9 combinations.

Fig. 2 summarizes the data as viewed with respect to both reproductive state and female density. "X" indicates the intensity of overall population aggregation; that of the individual components is indicated by symbols in the corners. Among non-gravid females (A), it is evident that population density decreased with female density. When only a few non-gravid females were present, male aggregation played an important part in overall aggregation. Otherwise, the populations tended to show weak aggregation, with that at 16♀ the only group which showed an absence of aggregation. Populations with gravid females behaved quite differently (B and C). In both, aggregation increased with increased female density. This reflected nymphal and female aggregation relationships; males did not aggregate to a detectable extent. A difference according to reproductive state was evident in that groups with immature egg cases (B) tended to aggregate more than those with older egg cases (C). This was primarily due to more intense aggregation among females. It is evident that the female reproductive state mediates the effects of density on the aggregation of the population as a whole and that it influences the aggregation behavior of the individual components of the populations.

It would require studies beyond those covered in the present contract to fully explore the relationships revealed above, especially as to whether and/or how these might be related to dispersion. Although we have laboratory data on dispersion, the experiments were not designed to investigate such relationships, for the simple reason they were hitherto unknown. It is noteworthy that all gravid females aggregated. This may be related to a general absence of gravid females from "peripheral" traps in the shipboard experiment (Obj. 3 - traps some distance from primary aggregation sites). At main aggregation sites, absence of gravid females from traps was clearly an effect of their feeding behavior (see Obj. 5). Within an area of limited harborage availability, surges in the numbers leaving a harborage, as seemed to occur in the shipboard experiment (Obj. 3), might correlate with periods of high density of non-gravid females.

Objective 3 - To analyze the behavior of cockroach populations

Laboratory experiment: This experiment used 3 inter-connected aquaria. Food, water, and shelter (black construction paper as in Obj. 2) were provided in the 2 end aquaria. Mixed populations, similar to those used in the within-harborage aggregation experiment, were released in one of the end aquaria. Movement into the adjoining aquaria was prevented for the 1st day. Subsequently, free access through plastic tubing was allowed. The cockroaches remaining in their initial location and those in each of the two adjoining



acquaria were censused at the end of one week. The data are being analyzed to determine: 1) the proportion of each component that left the original harborage, and 2) those that moved to establish a new aggregation in the other end aquarium. Basically, this experiment looks at a situation where an equally favorable harborage is located a short distance from an inhabited harborage. The data are in the process of statistical analysis. The results will be presented in a later report.

Shipboard experiment. Part I - Data from wild-type cockroaches

The experiment was conducted on an inactive subtender at the U.S. Navy Inactive Ship Facility, Portsmouth, Va. The area of study consisted of the galley and adjacent rooms (Fig. 3). Cockroaches used to establish populations were drawn from a wild-type strain collected from a Navy vessel and reared in the laboratory for two years.

The experiment was initiated with a survey in which patches of fecal matter were used to locate aggregation sites of previous infestations. Eleven of these were selected as sites of release (Fig. 3, 1-11). Dog food pellets were scattered around each release site; water was supplied in a plastic container with 4 cotton wicks extending through the lid into the water below. Roatel® traps were used to assess group establishment following the initial releases and later to conduct a summer-long trapping program.

Releases at each of the 11 sites were made on 4/14, 4/28, 5/12, and 5/27, 1981. Those on 4/14 and 4/28 consisted of 5 females with immature first egg cases (stage I, Tanaka 1976), 10 3rd to 4th stage nymphs (5♂, 5♀), and 10 1st to 2nd stage nymphs (5♂, 5♀). On 5/12 and 5/27, releases were increased by doubling the number of nymphs. Thus, a total of 20 gravid females and 120 nymphs (60♂, 60♀) were released at each site, giving a total release of 1,540 cockroaches. Success of group establishment was judged by visual survey and by leaving two traps onboard between each visit. Following negative observations on 4/28 and 5/12, the release procedure was altered. Instead of emptying the cartons in which cockroaches were transported, a carton was left at each site. Opportunity for the cockroaches to leave was provided by either loosening the lid or cutting a hole in the side. Releases were discontinued following sightings of cockroaches at or near release sites on 5/27.

Data were collected on relative humidity, using an Atkins Thermocouple Psychrometer. Temperature was also recorded at each visit. In addition, variation was determined by leaving a maximum-minimum thermometer on board between each release.

On 6/8, a trapping program was initiated. Traps were placed at each release site, henceforth referred to as "main" sites, and at various distances and locations from the main sites ("peripheral" sites) (Fig. 3). The initial data were from traps at the main sites, #1-11, and 9 peripheral sites, as listed in Table 3, col. 2. Peripheral traps were either numbered according to a main site or, if separated from main sites by large, open areas or walls, given separate numbers. For example, traps peripheral to site 1 are numbered as "1A" and "1B". During the morning of 6/9, catch of each trap was recorded

by noting the number of non-gravid adult females, gravid (egg case-bearing) females, adult males, middle to late stage nymphs, and small nymphs (primarily stages 1 and 2). After counting, cockroaches were given the opportunity to leave the traps by removing the lids. Later, the traps were tapped gently until no insects remained within.

On 6/23-24, trapping data were collected similarly to the above, but traps were put out for two successive nights. Also, the number of traps was increased, bringing the total to 39. On the next trapping data (7/14-15), the total number of traps was increased to 45, with locations as shown in Fig. 3. The program of two trap nights was continued at 3 wk intervals throughout the summer, with final trapping data collected on 8/25-26. A variation with respect to resource location was introduced at the time of the preceding trip, 8/4-5. Before leaving, food and water were moved from site #2 to a peripheral site, #2A, which was then provided with a shelter (carton). Also, food, water, and a carton were added to a corner galley site, #3C (Fig. 3).

Three types of collections were made at the conclusion of the experiment. Instead of releasing the catch from the 2nd night of trapping (8/26), traps with heavy catch were placed in plastic bags, sealed, and brought back to the laboratory for study. Likewise, each carton was collected, but not until we were ready to begin a final "cleanout" of the surrounding area. Cockroaches caught as a result of insecticide treatment constituted the third type of collection. Prior to treatment, open areas in the galley were ringed with sticky tape; in the small rooms, tape was used around the doorways. As in preceding studies, synergized pyrethrins spray was used to flush cockroaches, followed by spraying 1% propoxur in oil into the harborage (Keil 1981; Ross, Keil and Cochran, 1981). Spraying began at the outer margins of the area and progressed inwards towards the main harborage. Collections were made with a modified vacuum cleaner and by hand and the insects preserved in 70% alcohol in appropriately labeled bottles. A team of 6 worked the area around each of the main sites for approximately 20-25 min. Four then went on to prepare the next area (remove carton; ring with sticky tape), while two remained behind to collect any additional cockroaches that came out of the harborages. When this process had been completed for all areas, each was again surveyed for remaining cockroaches, including those caught on sticky tape.

Groups at each site were established in as uniform a manner as possible. Nevertheless, groups developing at these sites throughout the summer showed marked differences in their characteristics. These differences were related to their immediate surroundings. Therefore, a brief description of the major features of each site is given below (locations in Fig. 3):

- #1 - isolated from other cockroaches and resources by distance and galley wall; surrounded on 3 sides by large open spaces.
- #2 - corner site, with large vertical pipe and extensive loose flagging.

#s 3 & 8 - under serving lines; protected routes of movement along pipes under serving lines.

#s 4 & 7 - under equipment separated from galley wall by space.

#5 - adjacent to wall that was decayed extensively.

#s 6, 9 & 11 - under comparatively high, open shelves.

#10 - under low-lying equipment adjacent to wall.

The analysis of the growth and behavior of wild-type groups is partially completed. This information is presented below. We are currently studying changes in age class structure as these occurred throughout the summer at each main site and at related peripheral sites. We expect these results will be ready for inclusion in our 6 mo report (Aug., 1982).

a. Establishment of wild-type groups. - Difficulty was encountered in establishing wild-type groups. The first visual sightings (site #2, 5/27) followed the onset of more consistently warm weather in late May. An additional factor in group establishment may have been the change in procedure whereby cockroaches were left in the cardboard cartons in order to limit initial dispersal (a suggestion of LCDR McCroddan, for which we thank him). Presence of cockroaches at the main sites was confirmed by catch at each of the main sites on 6/9.

b. Trapping data according to general location. - Table 3 shows trapping data according to main and nearest peripheral trapping sites; Table 4 shows those from traps separated from main sites by either large open spaces or by a wall (locations in Fig. 3). Most of the cockroaches were caught at the main sites, indicating groups were established near food and water, either in the cartons or nearby harborages afforded by the ship. However, continued growth of these groups was accompanied by increased catch in the nearest peripheral traps (Table 3, sites marked by "A", "B", or "C"). Sudden sharp increases on 8/25-26 in certain peripheral traps previously characterized by little or no catch, such as 1A and 1B, resulted from re-location of food and water (see below). The last set of trapping data also showed higher catch from the 2nd night of trapping (2,110 vs 1,606 on 8/25). This resulted from more accurate counts of small nymphs in traps brought back to the laboratory than in visual surveys onboard (small nymphs sometimes hidden in bait or corners and difficult to see). From Table 4, it is clear that open spaces (#s 13-15) and walls (#s 16-28) acted as barriers to movement.

The totals trapped throughout the summer show a steady increase (Table 3, last line). Within the 3 mo period between 6/23-24 and 8/25-26 (= one generation), the overall population(s) increase was 18.6x.

c. Trapping according to specific sites. - Comparative catch at the main sites indicated differences in the density of major aggregations as well as in their rate of growth throughout the summer (Table 3). These reflected

differences in their immediate surroundings, as noted above. Large groups developed where loose pipe flagging, decayed walls, or low-lying equipment next to a wall provided narrow, hidden recesses close to main sites (#s 2, 5 and both 1 and 10, respectively). In late June, the two largest groups were apparently at sites #1 and #2, with development at #1 possibly reflecting isolation as well as secluded harborages. It was not until late summer that the group near #5 became the 3rd largest. This reflected the discovery and infestation of the decayed wall at #5 (Fig. 3). It was such a favorable site that cockroaches in the carton at #5 apparently moved into the wall (note comparatively low numbers in carton at #5 in Table 5, next to last col.). Lowest catch at main sites was associated with high, open shelves (#s 6, 9 and 11) or under equipment separated from the main galley wall (#s 4 and 7).

The rate of growth of groups at the main sites reflected the differences in their immediate surroundings, with an added factor of movement from certain less favorable spots towards locations where large infestations were developing. Among the 4 largest groups, those at #s 1 and 2 showed marked increases in June (Fig. 4A and B); in contrast, those at #s 5 and 10 showed little growth until after mid-July (Fig. 4A and 5A, 3rd set of trapping data). Increases at #s 5 and 10 almost certainly reflected additions from cockroaches that left groups established at less favorable sites, as well as growth within groups already at #s 5 and 10. A comparison of growth at the two bakery sites, #s 10 and 11, suggests the group at #10 grew at the expense of that at #11 (Fig. 5A). That at #5 probably contained migrants from #s 9 and 7, using a route shown by black-body during late summer (from #9 to #7 towards 7B and into the wall at #5) (Shipboard experiment: Part II). A "wave" of cockroaches leaving #9 and passing through #7 could account for the simultaneous decrease at #9 and increase at #7 seen in trapping data from 8/4-5 (Fig. 6B, trap set 4). Among the 4 largest groups, that which started with largest catch (#2 on 6/9) outgrew the other three up until removal of food and water on 8/5 caused a sharp decrease (Fig. 4, 4th trap set).

Low catch throughout most of the summer indicated the serving lines were not particularly favorable sites for the establishment of large aggregations (Fig. 5B, trap sets 1-3). Rather, increased catch at these sites during August, especially at #3, almost certainly reflected increased movement. That at #3 was markedly affected by re-location of resources (removal from #2; addition to both 2A and 3C) (see below). Data from trapping at the least favorable sites are shown in Fig. 6. Sharp increases in catch at #s 4 and 6 in the last set of trapping data may have reflected increased movement around the galley margins, both that occurring naturally (such as cockroaches leaving #s 9 and 7) and that stimulated by re-location of resources. Both increased density of and increased movement in the area around groups established at main sites resulted in increased catch. Least growth occurred among groups at #s 7 and 9 (Fig. 6B).

Fig. 4B shows the effect of removing food and water on the large group established at site #2. The precipitous drop at #2 was reflected in increases at the two nearest main sites (#1, Fig. 4A; #3, Fig. 5B). It also caused

sharp increases in peripheral traps in this general area (Fig. 7). Although food, water, and shelter were provided at 2A, there was more movement towards established groups (#s 1 and 3) than towards this previously uninhabited spot. However, more cockroaches were caught on the far side of #1 (1A) than in the trap lying between #1 and #2 (1B).

The above data document effects of resource availability that are difficult to demonstrate in natural populations since experiments usually have to be conducted where there is a lack of control and/or knowledge of the actions of human inhabitants. Large numbers left a favored harborage (#2) when it became necessary to forage over a greater distance for food and water. It appeared that the cockroaches were attracted to other groups, rather than to newly established harborages furnished with food and water (except for gravid females, see p.11). Resources were added at #3C as well as 2A, yet neither became the focus of a heavy group, at least within the 3 wks between the last two sets of trapping data (Table 3). The most surprising aspect of increases in the peripheral traps was the greater increase at 1A than 1B (Fig. 7A). Trap 1B lies between #2 and #1 (Fig. 3). It is as if cockroaches initially attracted towards the group at #1 were forced to move beyond #1 towards #1A. A similar pattern was seen in late summer movement patterns of black-body. This suggested a repellent effect associated with high density groups where favored harborage space is limited. These data from a natural population support a recent study by Suto and Kumada (Appl. Ent. Zool. 16: 113-120, 1981). They reported a dispersion-inducing substance in the saliva of adults that causes nymphal dispersion at high adult density. No distinction was made between adult males or females or, among the latter, according to reproductive state. It would be interesting to determine whether secretion of the dispersion-inducing substance does indeed vary among such adults and, if so, if it might have a relation to within-harborage aggregation as seen in Obj. 2. It may well represent the mechanism that causes nymphs to move away from established harborages, especially where these are in less favorable locations and cockroaches become crowded into limited spaces.

d. Final collections. - Table 5 summarizes data from collections made at the termination of the experiment. In general, the numbers caught (trapped) at each of the main sites (col. 3) corresponded reasonably well to the total collection from that site and the surrounding area (col. 2). At 7 of the 11 main sites, the catch fell within the range of 17-21% (exclusive of new sites, 3C and 2A). Higher % catch correlated with the two sites (#s 1 and 3) at which sudden increases in numbers followed removal of resources from #2. The influx of cockroaches also resulted in increased movement in the area around #s 1 and 3, causing very high % catch from these areas as a whole (col. 6). High % catch at the new site, 3C, also reflected movement; very few cockroaches were in the "new" harborage (carton) at that site (last col.). Lowest catch correlated with the location of groups that showed the least growth, i.e. #s 7 and 9.

Table 3 showed that the distance of a trap from a primary aggregation had a marked effect on the catch. The above data suggest two additional factors. Increased movement, especially that artificially stimulated by re-locating

food and water, increases the percent catch. In addition, catch near groups inhabiting less favored harborage areas (groups with little growth) was lower than that at other locations. Possibly there was a greater tendency of cockroaches in such situations to remain in the cartons rather than to move out into relatively open areas around the carton. Alternatively, catch might be related to the makeup of populations that constitute such groups (age class frequency data not yet fully analyzed).

The number of cockroaches in the cartons also showed a rough correlation with the total/area, primarily because the cartons generally remained favored harborages (Table 5). The major exception was at #5, where most of the cockroaches had moved into the nearby decayed wall. Among cartons that remained a major harborage within their areas, the lowest proportions were in those under the serving lines (#3 and 8), where protected routes of movement were available along horizontal pipes running under the serving lines. In contrast, the percent of cockroaches within cartons was usually high if the immediate surroundings offered little seclusion, as at #s 2A, 9, 7, 4 and 6 (Table 5, last col.). Abundant harborage was available at #2 and, in this case, it appears that cockroaches within the carton may have been slower to leave the area following removal of food and water than those outside the carton.

e. Notes on population structure of wild-type groups. - Data from the final collections have been summarized indicating population structure among cockroaches from three sources: cartons (= main harborage); traps at main harborages (adjacent to cartons); and collections made in the course of insecticide treatment around and including each main site.

A comparison of gravid vs non-gravid females from cartons and nearby traps showed a higher proportion of non-gravid females in the traps. A total of 520 non-gravid females was collected. Of these, 446 were in the traps (86%). Gravid females totalled 176. Of these, 48% were trapped. The data on cyclic feeding behavior (Obj. 5) provide a partial explanation of these differences. As expected, the data indicate gravid females have less need of food than non-gravid females. In addition, it appears the roatel® bait was highly attractive to females, but only over a short distance. A disproportionate number of females was found in the traps in comparison with catch of other components of the population, with the exception of small nymphs. Thus, the data from the traps showed approximately a 2:1 ratio of females to males; yet, if the data from the main sites (carton and nearby trap) are added together and compared to those from the cleanout collection, it is clear there was an overall 1:1 sex ratio, as seen below:

<u>Collection</u>	<u>Ad.♂</u>	<u>Ad.♀</u>	<u>Nymphs<sup>a</sup></u>	<u>Total</u>
Carton	364	153	562	1,079
Trap	351	627	509	1,487
Cleanout	589	592	1,286	2,467

<sup>a</sup>Middle to late stage nymphs only.

The collections from the main site (carton & trap) show somewhat larger numbers of adults than those of the cleanout. The latter contained the largest number of middle to late stage nymphs. Possibly this was a natural situation reflecting a tendency of the nymphs to be more widely spread around a main harborage and/or to disperse from the harborage; alternatively, it might reflect a wider dispersal of nymphs following insecticide treatment, as seemed to be the case in the sterile male experiment. More data on this aspect are expected from the 1982 summer experiment.

Within the carton-type harborage, small nymphs formed a very high proportion of the population (avg. 79%; range from 71-86%). Like the gravid females, they tended to remain within the harborage. Within those traps that were brought back to the laboratory for analysis of contents (2nd nights trapping), small nymphs averaged 46% of the total collected.

Shipboard Experiment. Part II. Data from the introduction of mutant markers

The data analyzed thus far were largely from trapping. They were summarized in the proposal for the current Contract year (1982-1983). Rather than to simply repeat those data, we list here a few of the major points:

1. Hatch of successive egg cases and the rate of nymphal development as seen from the trapping data were closely similar to those observed in the laboratory.
2. Gravid females joined wild-type groups at or near the site at which they were introduced (black-body at #9; orange-body 3 wks later at #10).
3. Hatch of 1st, 2nd, and, in the case of black-body, 3rd oothecae, indicated the parental females generally remained in their initial locations (black-body around #s 9 and 7; orange-body at #10), as did most of the small nymphs.
4. In contrast, many middle to late stage nymphs dispersed from the main sites where they had hatched. Black-body showed routes of movement from #s 7 and 9 around the margins of the galley, with that towards #s 4 and 5 favored over that towards #s 8 and 6 in late summer (route apparently followed by wild-type leaving #9 and passing through #7 towards #7B and eventually the decayed wall at #5 in late summer). Possibly the change in the preferred route of movement reflected an attraction towards the large aggregations that became established at #5 in late summer. An attraction towards established groups was evident in wild-type data following removal of food and water from #2.
5. Movement of older nymphs was continued into adult stages, but once females became gravid, they joined groups at main sites (near food and water).
6. Each set of middle stage nymphs from black-body females was more widely dispersed than that of the preceding set (hatch from preceding oothecae). This may be added evidence of a repellent effect of groups at high density for their particular harborage, as suggested by wild-type cockroaches that left #2 and passed beyond #1 towards 1A.
7. Orange-body showed patterns similar to black-body. By far the largest collection of small nymphs from 2nd egg cases was in cartons left at #10, the site at which gravid females had been released 6 wks earlier (215 1st

to 2nd instars at #10; 69 at other bakery site, #11; and 10 divided among 4 different cartons in the galley). In contrast, more mid to late stage nymphs had left the bakery and moved into the galley. However, fewer had left the bakery in comparison to black-body leaving the area of #s 7 and 9, possibly because the bakery walls restricted movement.

At the end of summer, the hatch of a 2nd generation of black-body had begun. It is interesting that these nymphs occurred in cartons from all main sites, although with marked differences in number. Most, but not all such nymphs were hybrids, showing matings between F<sub>1</sub> black-body and wild-type. Cartons with the largest numbers were from: #7, where the parents had hatched and spent their life span; #4, on the route followed by black-body nymphs in late summer; and, surprisingly, from #2A. Undoubtedly, these were among the cockroaches that left #2 following removal of food and water. The carton collected from 2A was almost entirely populated by adult females and newly hatched nymphs, both black-body and wild-type. A higher proportion of the former moved to 2A; overall, the largest numbers of wild-type leaving #2 moved towards #s 1 and 3, as noted above. However, wild-type with greatest need of water, i.e. gravid females and small nymphs from #2, were apparently differentially attracted to the new source at 2A, even though other components of the population moved towards #s 1 and 3.

#### Objective 5 - Foraging habits

It was reported earlier that female feeding behavior was studied over the reproductive life of a group of individuals. Corresponding studies are now complete with respect to drinking behavior. Again, here the objective was to determine female requirements during reproduction which may relate to foraging behavior.

A series of large female nymphs were set up in individual cages provided with food and a small harborage. Water was offered daily for a period of one hour. The watering device was weighed at the beginning and end of this period. It was necessary to restrict water availability in this manner because loss from evaporation over a 24 hour period was so large in relation to the amount drunk that the latter could not be determined accurately. Even in one hour, a correction factor must be introduced to allow for evaporation.

The nymphs were observed daily and days on which maturation, mating, egg case production and egg case hatch occurred were noted. The adults were followed through production of the fourth egg case.

The results indicate that, as with food, there is a clear cyclic drinking pattern. After hatch of an egg case, females drink large volumes of water until just prior to formation of the next egg case. Subsequently, they drink sparingly, taking a drink perhaps every third or fourth day while carrying an egg case. The drinking pattern prior to formation of the first egg case is a little different. Newly-emerged adult females tend to drink periodically more like nymphs. About the time they are ready to mate, they begin to drink regularly and continue to do so until just prior to formation of the first egg case.



Thus, it appears that reproducing German cockroach females eat and drink cyclically. While adult females in an infestation population are not likely to be reproducing synchronously, these results point out that reproducing females may be especially susceptible to food and water deprivation during certain times in their reproductive cycle.

Publications of research supported in part by this Contract

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Ms. in preparation:

Bret, B. L. and M. H. Ross. Laboratory experiments on within-harborage behavior of the German cockroach.

Major invited talks

1980 - Sterile male experiment was described as part of a symposium paper at the XVth International Congress of Entomology, Kyoto, Japan. (Ross).

1981 - The shipboard experiment will be described in a symposium paper to be presented at the forthcoming national meeting of the Entomological Society of America, San Diego. (Bret).

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We owe a special debt of gratitude to those who made possible the summer "field" experiment on the inactive ship. Access and lighting arrangements were made through the cooperation of the Inactive Ship Maintenance Facility at Portsmouth. Captain H. J. Candela, Navy Environmental Preventive Medicine Unit #2, appointed LCDR Donald M. McCroddan as our liason officer, and with his good help and that of those under his direction, especially H.M. 1 Slinker, Chief Francis, and HM 2 Julia Tettaton, we were able to proceed with the summer's work.

TABLE 1 - RESULTS SUMMARIZED ACCORDING TO DENSITY  
OF THE FEMALES

4♀			.43	S
		.48	.45	L
	.35	.41	.38	♂
.42	.44	.42	.43	♀
♀	♂	L	S	.44

8♀			.43	S
		.36	.40	L
	.30	.30	.32	♂
.36	.33	.33	.38	♀
♀	♂	L	S	.36

16♀			.60	S
		.45	.51	L
	.29	.31	.32	♂
.41	.32	.43	.48	♀
♀	♂	L	S	.44

TABLE 2 - RESULTS ARRANGED BY REPRODUCTIVE STATE  
OF FEMALES

A				.44	S
			.41	.41	L
		.35	.37	.34	♂
	.25	.31	.32	.31	♀
	♀	♂	L	S	.36
B				.53	S
			.47	.50	L
		.32	.36	.33	♂
	.58	.38	.47	.54	♀
	♀	♂	L	S	.47
C				.49	S
			.40	.44	L
		.26	.30	.34	♂
	.39	.32	.40	.45	♀
	♀	♂	L	S	.41

Table 3. Summary of wild-type trapping data.

Trap location <sup>a</sup>	Date and number caught						Total
	6/9	6/23 - 6/24	7/14 - 7/15	8/4 - 8/5	8/25 - 8/26 <sup>b</sup>		
#1	3	13 - 17	27 - 52	64 - 100	196 - 249 <sup>c</sup>		721
#1A	-	0 - 0	1 - 3	15 - 6	97 - 147 <sup>c</sup>		269
#1B	-	0 - -	2 - 0	0 - 1	36 - 83		122
#2	15	30 - 12	124 - 74	226 - 145	94 - 81		801
#2A	-	0 - 0	0 - 0	0 - 0	43 - 58 <sup>c</sup>		101
#3	5	3 - 4	20 - 4	65 - 75	122 - 183 <sup>c</sup>		481
#3A	-	0 - 2	0 - 0	15 - 13	89 - 84		201
#3B	-	0 - 0	7 - 5	16 - 25	80 - 118 <sup>c</sup>		251
#3C	0	1 - 1	1 - 3	33 - 26	71 - 87 <sup>c</sup>		223
#4	5	6 - 3	15 - 12	5 - 17	54 - 60		177
#5	2	2 - 3	26 - 28	66 - 78	151 - 200 <sup>c</sup>		556
#5A	-	0 - 1	4 - 9	19 - 24	83 - 153 <sup>c</sup>		293
#6	2	1 - 4	14 - 3	10 - 11	26 - 57		128
#6A	0	1 - 0	1 - 0	4 - 5	10 - 12		33
#7	8	7 - 16	24 - 8	38 - 31	20 - 15		167
#7A	1	4 - 3	16 - 8	15 - 6	30 - 40		123
#7B	1	3 - 1	12 - 14	19 - 21	71 - 48		190
#8	5	5 - 6	13 - 19	43 - 44	36 - 64 <sup>c</sup>		235
#8A	-	0 - 0	1 - 0	0 - 0	11 - 11		23
#8B	2	7 - 5	1 - 1	5 - 5	25 - 13		64
#8C	1	4 - 1	2 - 1	8 - 15	40 - 11		83
#9	2	7 - 7	14 - 30	14 - 9	20 - 19		122
#9A	-	1 - 3	2 - 4	7 - 3	12 - 15		47
#10	2	4 - 4	28 - 34	39 - 49	110 - 173		443
#10A	2	0 - 0	0 - 0	1 - 2	2 - 1		8
#10B	-	0 - 0	0 - 0	2 - 0	11 - 14		27
#11	11	2 - 6	17 - 8	22 - 23	50 - 110 <sup>c</sup>		249
#11A	0	0 - 0	0 - 0	7 - 3	16 - 13		39
	67	101	99	372	320	758 737 1606 2119	6179

<sup>a</sup>Main sites = #1 - 11; peripheral traps numbered in respect to main sites (see Fig 1).

<sup>b</sup>Catch at #s 1, 2, 3, and adjacent traps influenced by relocation of food and water (see text)

<sup>c</sup>Brought back to the laboratory for counting (8/26).

Table 4. Peripheral traps that were separated from main sites by either large open spaces or by walls.

Trap location	Date and number trapped			
	6/23-24	7/14-15	8/4-5	8/25-26
#13	0 - 0	0 - 1	1 - 0	3 - 0
#14 <sup>a</sup>	0 - 1	0 - 0	1 - 2	2 - 0
#15	0 - 0	0 - 0	0 - 0	1 - 1
#16	0 - 0	0 - 0	1 - 3	0 - 0
#17	0 - 0	0 - 0	1 - 0	2 - 0
#18			0 - 0	0 - 0
#19			0 - 0	0 - 0
#20			1 - 0	0 - 0
#21	0 - 0	0 - 0	0 - 1	0 - 0
#22	0 - 0	0 - 0	0 - 0	1 - 1
#23		0 - 0	0 - 0	0 - 0
#24				3 - 6
#25	0 - 0	0 - 0	0 - 0	0 - 1
#26	0 - 0	0 - 0	0 - 0	0 - 1
#27				14 - 8
#28		0 - 0	0 - 0	1 - 1

<sup>a</sup>Only trap in this list that was put out on 6/9. One cockroach was caught.

Table 5. Data from collections made at the termination of the experiment (8/25-28).

Area <sup>a</sup>	Total/ area <sup>b</sup>	Trapped at main site:		Total trapped/ area		In carton:	
		No.	%	No.	%	No.	%
#5	1,903	350	18	586	31	290	15
#10	1,433	283	20	311	22	816	57
#1	1,243	445	36	808	65	709	57
#3	1,112	305	27	676	61	465	42
#3C	567	158	28			78	14
#11	954	160	17	189	20	493	52
#2	944	175	19			777	78
#2A	865	101	12			745	86
#9	700	39	6	66	9	473	68
#7	660	35	5	224	34	437	66
#4	535	114	21			417	78
#8	466	100	21			114	25
#6	414	83	20	105	25	327	79

<sup>a</sup>Area listed according to main site within that area. Nos. 3C and 2A are included because of resources placed at these locations on 8/5.

<sup>b</sup>Total/area = numbers of cockroaches removed in traps (2nd night's catch, 8/26) plus those in carton and from cleanout collections.



FIG. 1 - PERCENTAGE OF THE COMPONENTS OF POPULATIONS AMONG FOUR "RE-NUMBERED" SHELTERS WITH RESPECT TO FEMALE DENSITY

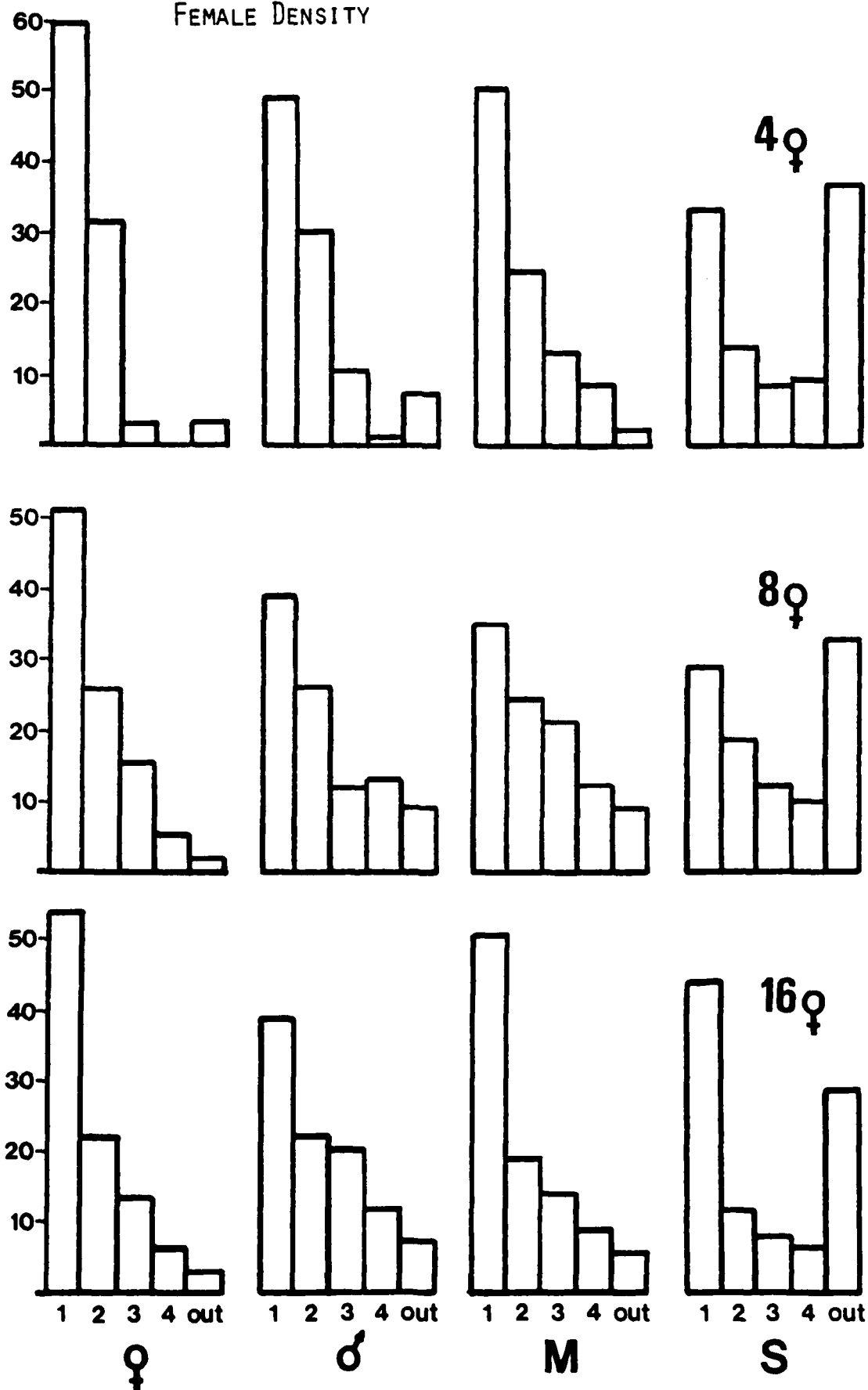
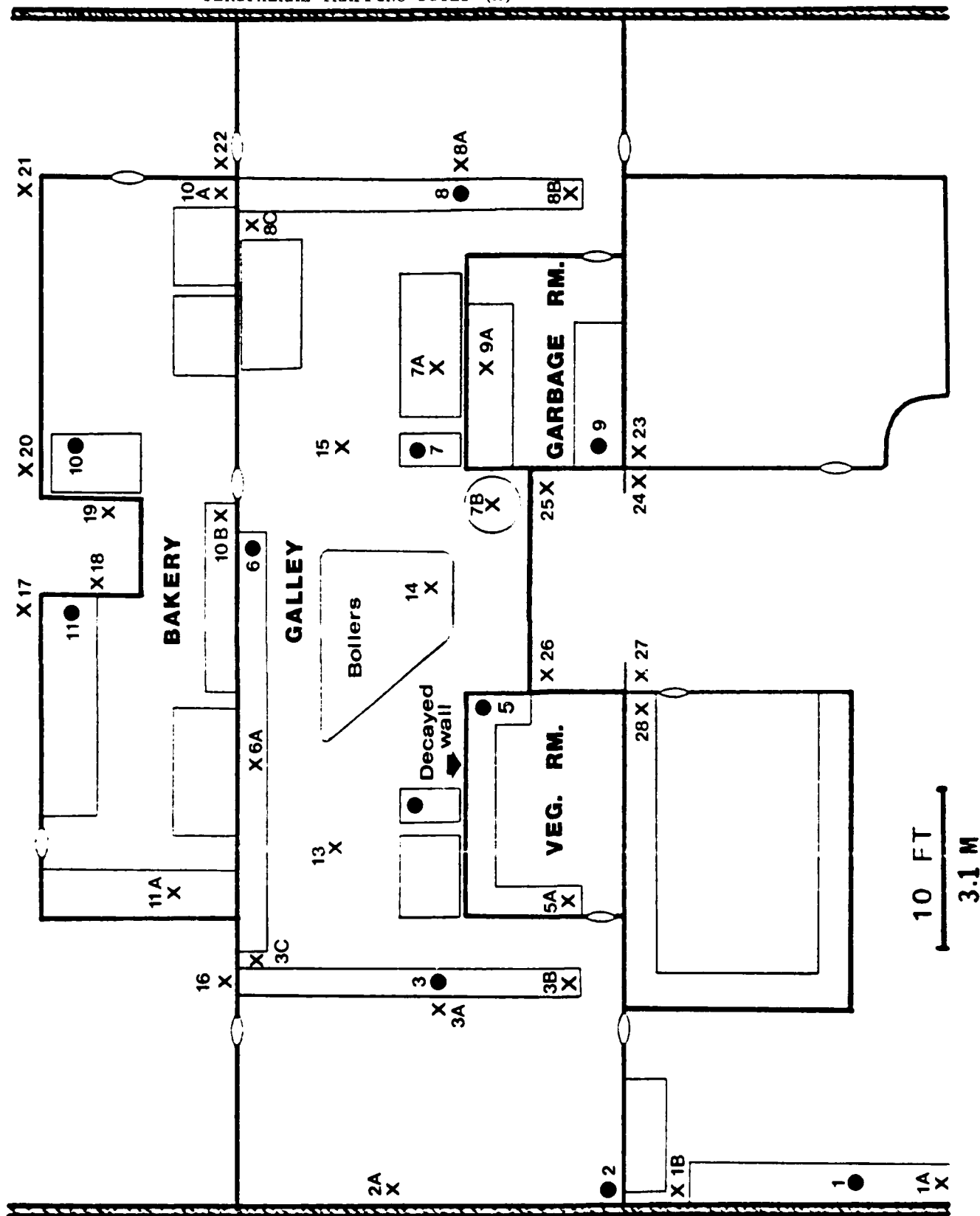


FIGURE 2. SUMMARY OF WITHIN-HARBORAGE RELATIONSHIPS

(A - females without oothecae; B - females with immature egg cases (stage I); C - females with advanced egg cases (stage IX-X)).

	A		B		C	
4♀	♀♀ XXX MMM	SSS — SSS	♀♀ XX MM	— — SS	♀ X M	— — SS
8♀	— X M	— — SS	♀♀ XX MM	— — SS	♀♀ X MMM	— — SSS
16♀	— — M	— — SS	♀♀♀ XXX MMM	— — SSSS	♀♀ XX MMM	— — SSS

FIGURE 3 - MAP OF THE EXPERIMENTAL AREA SHOWING LOCATIONS OF MAIN SITES (●) AND PERIPHERAL TRAPPING SITES (X)



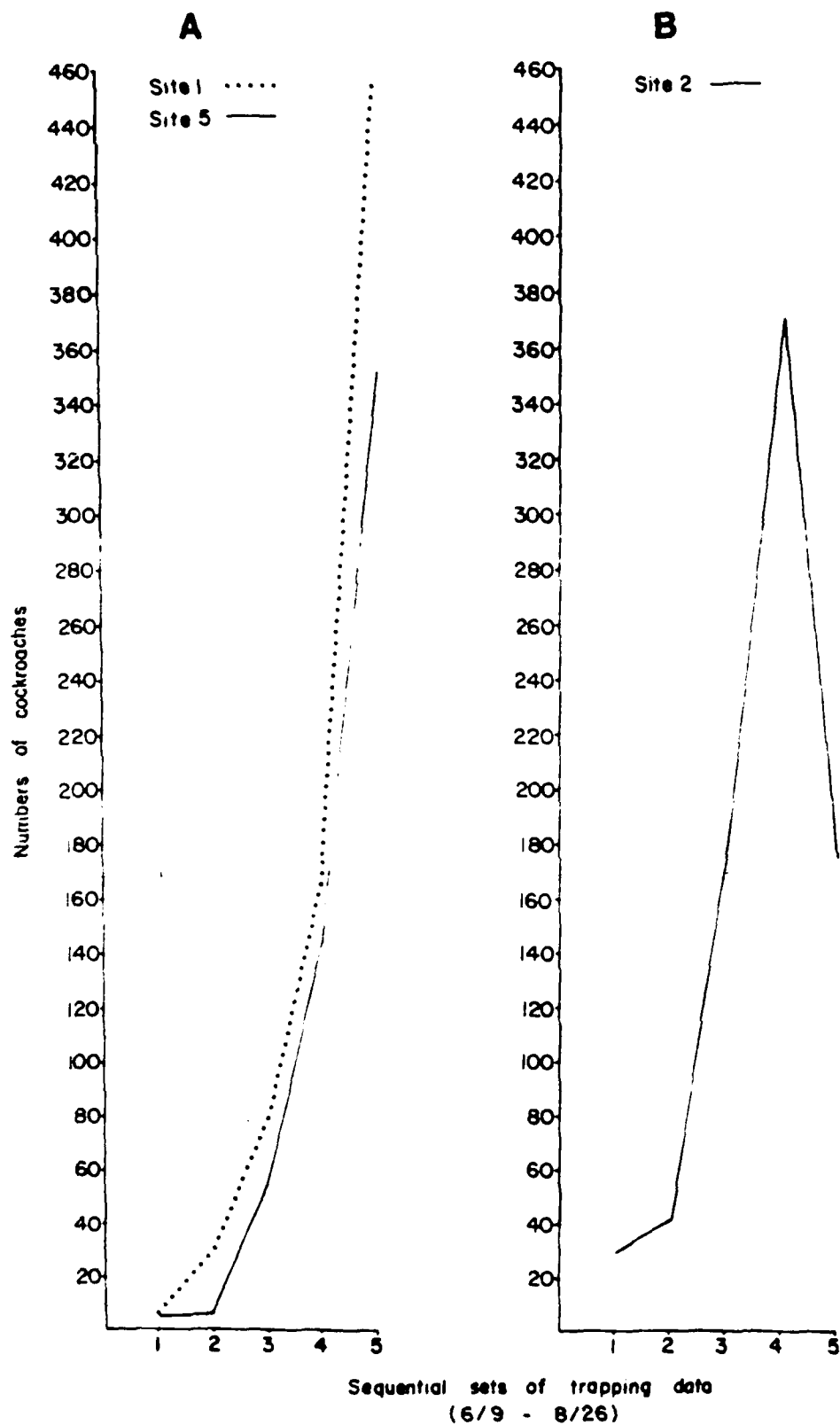


FIGURE 4 - GROWTH OF GROUPS AT FAVORABLE HARBORAGE SITES

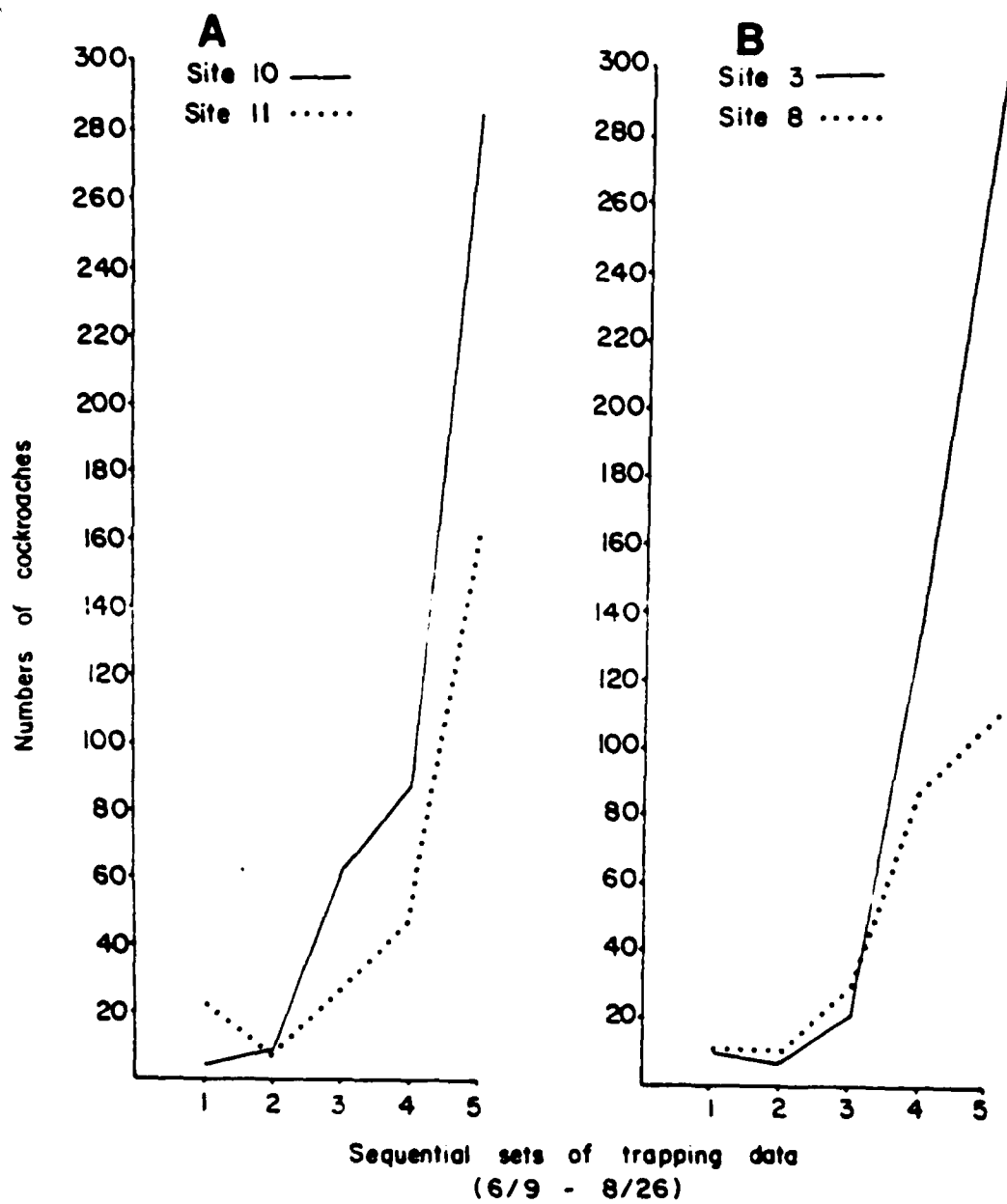


FIGURE 5 - GROWTH OF GROUPS AT THE TWO BAKERY SITES (A) AND THE SERVING LINES (B)

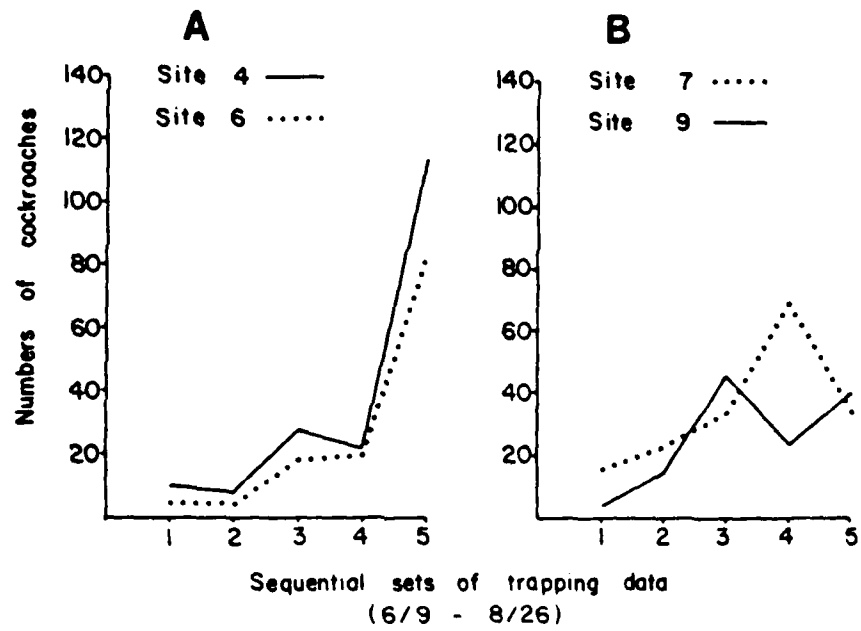


FIGURE 6 - GROWTH OF GROUPS AT LEAST FAVORABLE SITES

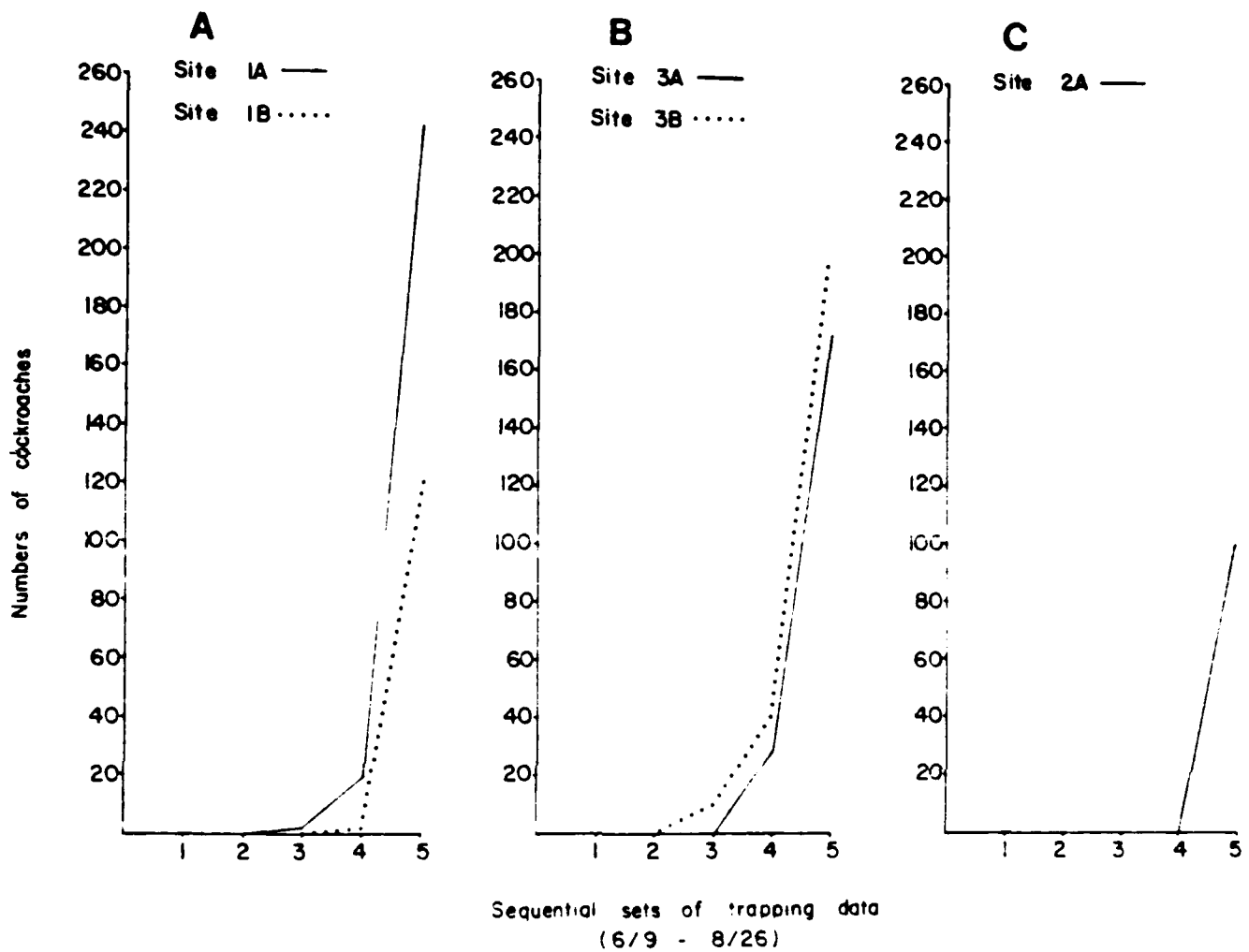


FIGURE 7 - EFFECT OF REMOVING FOOD AND WATER FROM #2 FOLLOWING 4th TRAPPING (8/5) ON CATCH IN NEAREST PERIPHERAL TRAPS ON 5th AND FINAL SET OF TRAPPING DATA (8/25-26)

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